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# Relationship of Dietary Iodide and Drinking Water Disinfectants to Thyroid Function in Experimental Animals

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**The importance of dietary iodide on the reported hypothyroid effect of drinking water disinfectants on thyroid function was investigated. Previous studies have also showed differences in the relative sensitivity of pigeons and rabbits to chlorinated water. Pigeons and rabbits were exposed for 3 months to diets containing high (950 ppb) or low (300 ppb) levels of iodide and to drinking water containing two levels of chlorine. Results showed that the high-iodide diet prevented the hypothyroid effect observed in pigeons given the low-iodide diet and chlorinated drinking water. Similar trends were observed in rabbits exposed to the same treatment; however, significant hypothyroid effects were not observed in this animal model.**

**The factor associated with the observed effect of dietary iodide on the chlorine-induced change in thyroid function is unknown, as is the relative sensitivity of rabbits and pigeons to the effect of chlorine. Several factors may explain the importance of dietary iodide and the relative sensitivity of these species. For example, the iodine formed by the known reaction of chlorine with iodide could result in a decrease in the plasma level of iodide because of the relative absorption rates of iodide and iodine in the intestinal tract, and the various types and concentrations of chloroorganics (metabolites) formed in the diet following the exposure of various dietary constituents to chlorine could affect the thyroid function. The former factor was investigated in the present studies. Results do not confirm a consistent, significant reduction in the plasma level of iodide in rabbits and pigeons exposed to chlorinated water and the low-iodide diet. The latter factor is being investigated.**

## Introduction

Several investigators have observed hypothyroidism in experimental animals exposed to drinking water containing the disinfectant chlorine (pH 8.5), chlorine dioxide, or monochloramine (*1*, *2*). The concentration of the disinfectant used in one study to produce the hypothyroid effect in pigeons (white Carneau) was within the range observed in some municipal drinking water supplies (*2*). The latter study (*2*) suggests that drinking water disinfectants may pose a human health problem. However, the hypothyroid effect reported in pigeons was not observed in rabbits exposed to these three drinking water disinfectants at similar concentrations. The factors associated with the relative thyroid sensitivity of these animal models to the disinfectants is presently unknown. The level of iodide in the diets fed to the pigeons and rabbits may explain these differences. For example, the level of iodide in the pigeon feed was 300 ppb, whereas iodide levels of 950 ppb were measured in the rabbit feed. It is well documented that chlorine will oxidize iodide to molecular iodine (*3*), which can react with organic matter and give rise to iodoorganic compounds (*4*). The formation of these compounds may reduce iodide in the intestinal tract and thereby reduce the intestinal absorption and plasma level of iodide. A decrease in the level of plasma iodide can promote hypothyroidism. Thus an increase in the dietary level of iodide may prevent the hypothyroid effect observed in pigeons exposed to relatively low levels of iodide.

To determine the importance of dietary iodide on thyroid function, rabbits and pigeons were exposed to the disinfectant chlorine and diets containing relatively high (950 ppb) and low (300 ppb) levels of iodide. At 1-month intervals for 3 months, the plasma levels of cholesterol, 3′,5′,3,5-tetraiodothyronine ($T_4$), and free and bound iodine were measured. Cholesterol was determined in these studies because hypercholesterolemia is frequently observed in patients with hypothyroidism. Also, previous studies have shown increases in plasma cholesterol in pigeons receiving chlorinated drinking water.

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**Table 3. Effect of chlorinated drinking water and dietary iodine on plasma cholesterol, $T_4$, and iodine in pigeons exposed for three months.[a]**

| Plasma level | Diet | Control (deionized water) Zero time | Control (deionized water) 3 months | Chlorine (15 ppm, pH 6.5) Zero time | Chlorine (15 ppm, pH 6.5) 3 months | Chlorine (15 ppm, pH 8.5) Zero time | Chlorine (15 ppm, pH 8.5) 3 months |
|---|---|---|---|---|---|---|---|
| Cholesterol, mg/dL | Low iodide | 250 ± 12 | 246 ± 14 | 244 ± 21 | 295 ± 10 | 197 ± 17 | 281* ± 19 |
| | *High iodide* | 283 ± 31 | 278 ± 27 | 312 ± 19 | 310 ± 27 | 262 ± 17 | 281 ± 18 |
| $T_4$, (μg/dL) | Low iodide | 2.89 ± 0.15 | 2.60 ± 0.41 | 2.48 ± 0.23 | 1.80 ± 0.23 | 2.51 ± 0.34 | 1.30† ± 0.15 |
| | High iodide | 2.76 ± 0.38 | 2.90 ± 0.29 | 2.76 ± 0.90 | 2.80 ± 0.38 | 2.87 ± 0.18 | 3.34 ± 0.36 |
| *Iodine, μg/dL* | | | | | | | |
| Free | Low iodide | 0.97 ± 0.18 | | 1.29 ± 0.62 | | 1.86 ± 0.60 | |
| | High iodide | 1.83 ± 0.13 | | 2.05 ± 0.31 | | 3.94 ± 0.83* | |
| Bound | Low iodide | 4.67 ± 0.88 | | 3.04 ± 1.54 | | 3.27 ± 0.33 | |
| | High iodide | 3.46 ± 0.38 | | 5.06 ± 0.79 | | 3.46 ± 0.29 | |
| Total | Low iodide | 5.59 | | 6.35 | | 5.13 | |
| | High iodide | 5.39 | | 5.09 | | 7.40 | |

[a] *Mean ± SEM for four to five animals per experimental group. $p < 0.05$ and $p < 0.01$ were calculated by comparing within each treatment* group the zero-time and 3-month values (i.e., plasma cholesterol and $T_4$) or by comparing the controls and experimental groups at 3 months of exposure (i.e., plasma iodine).

*$p < 0.05$.

†$p < 0.01$.

were not significant. The only significant change observed for plasma iodine was an increase in the level of free iodine in the group receiving diet B and chlorinated water at pH 8.5.

The results thus far suggest that dietary iodide in the pigeon may alter the effect of chlorine on the thyroid function. In an attempt to explain this observation, the effect of chlorine (pH 6.5 and 8.5) and two other commonly used disinfectants (chlorine dioxide and monochloramine) on the release of bound iodide from diet was investigated. It has been suggested that iodide is absorbed through the intestinal tract at twice the rate of iodine. Chlorine is known to react chemically with iodide, giving rise to iodine, and is also thought to react with iodoorganics, giving rise to a chloroorganic and iodine. The reaction of chlorine with free and bound iodide may increase the dietary level of iodine and thus reduce the relative absorption of iodide with a subsequent change in thyroid function. Results shown in Table 4 suggest that these disinfectants, when added to the diet, reduce the level of bound iodide. Significant changes in bound iodide were not observed when the relative level of iodide was high in both the pigeon and rabbit diets. The greatest change was observed in the pigeon diet at low and high iodide levels when chlorine (pH 8.5), chlorine dioxide, or monochloramine was added at a concentration of 15 ppm. In the high-iodide rabbit diet, the addition of 2 ppm resulted in higher levels of bound iodide.

**Table 4. Effects of drinking water disinfectants on bound iodine in diets of pigeons and rabbits.[a]**

| Treatment group | Low iodide diet, μg/kg Pigeons | Low iodide diet, μg/kg Rabbits | High-iodide diet, μg/kg Pigeons | High-iodide diet, μg/kg Rabbits |
|---|---|---|---|---|
| Controls | 146 ± 18 | 176 ± 20 | 218 ± 29 | 272 ± 30 |
| Chlorine (pH 6.5) | | | | |
| 2 ppm | 133 ± 15 | 141 ± 18 | 238 ± 40 | 327 ± 24 |
| 15 ppm | 101 ± 8 | 107 ± 24 | 231 ± 28 | 243 ± 38 |
| Chlorine (pH 8.5) | | | | |
| 2 ppm | 131 ± 18 | 134 ± 19 | 197 ± 15 | 291 ± 27 |
| 15 ppm | 92 ± 7* | 110 ± 28 | 181 ± 17 | 240 ± 33 |
| Chlorine dioxide | | | | |
| 2 ppm | 149 ± 15 | 126 ± 10 | 202 ± 19 | 292 ± 35 |
| 15 ppm | 91 ± 9* | 82 ± 13† | 185 ± 17 | 247 ± 17 |
| Monochloramine | | | | |
| 2 ppm | 110 ± 15 | 148 ± 22 | 195 ± 16 | 321 ± 39 |
| 15 ppm | 64 ± 10* | 119 ± 13 | 191 ± 19 | 325 ± 26 |

[a] Mean ± SEM of two experiments per diet and per concentration of disinfectant. The ingredients in the low- and high-iodide diets are shown in Table 1. The procedure for measuring bound iodine and the effect of these disinfectants on the release of bound iodine are discussed in the Materials and Methods section. The concentrations of total iodine in the low- and high-iodide diets were 305 ± 29 and 956 ± 100 and 288 ± 36 and 985 ± 93, for pigeons and rabbits, respectively.

*$p < 0.05$.

†$p < 0.01$.

## Discussion

The effect of dietary iodide and drinking water disinfectants on thyroid function of rabbits and pigeons was investigated. It was apparent that dietary iodide in the pigeon influenced the effect of chlorinated water on thyroid function. Pigeons given the low-iodide diet and chlorinated water exhibited significant changes in plasma cholesterol and $T_4$. However, significant changes in these plasma constituents were not observed in pigeons fed the high-iodide diet and chlorinated water. In rabbits exposed to the low-iodide diet and chlorinated water, insignificant changes were observed for plasma cholesterol, $T_4$, and bound iodine. However, we did observe that high dietary iodide in rabbits prevented the small changes observed in the low-iodide group exposed to the chlorinated water, which suggests that statistical significance may be achieved with a larger number of experimental animals.

The results of these studies suggest that chlorinated

water may induce hypothyroidism in pigeons as evidenced by significant changes in plasma $T_4$ and cholesterol. The plasma concentration of cholesterol is significantly elevated in clinical hypothyroidism and generally bears a reciprocal relationship to the level of thyroid activity (*12–14*). However, the existence of similar reciprocal relationships in pigeons remains to be established. Nevertheless, we observed in previous (*15, 16*) and the present studies that a correlation exists between plasma cholesterol and thyroid activity. Furthermore, present studies suggest that dietary iodine may alter the effect of chlorine on both plasma cholesterol and $T_4$.

The relationship of chlorine to thyroid activity is not known. However, the fact that relatively high dietary levels of iodine prevented the significant effect of chlorine on plasma cholesterol and $T_4$ suggests that this disinfectant may affect iodine metabolism when the dietary level of iodide is relatively low. It has been shown that chlorine reacts with iodide, giving rise to iodine, an active oxidizing species (*17, 18*). Iodine in the presence of organic matter would be expected to react and give rise to iodoorganics. Chlorine is also thought to react with iodoorganics, affecting the release of iodide and possibly the substitution of chlorine at the iodide-vacated site. A decrease in the level of dietary iodine could reduce the plasma level of iodide and thus the activity of the thyroid because iodide is absorbed through the intestinal tract at a rate greater than that of iodine. In an attempt to determine the validity of this assumption, the effect of drinking water disinfectants on the release of bound iodide in the rabbit and pigeon diet was investigated. Results showed that these disinfectants reduce the level of bound iodide in the diet, the magnitude depending on the concentration of the disinfectant and the dietary level of iodide. For example, a significant decrease in the level of bound iodide was observed when the concentration of the disinfectant was increased from 2 to 15 ppm. However, this effect was only observed when the low-iodide diet was used, which suggests that the dietary level of iodide may influence the effect of these disinfectants on the release of bound iodide in the diet.

These *in vitro* studies suggest that these disinfectants reduce the level of bound iodide in the diet. If these effects occur *in vivo*, the dietary levels of iodide may be reduced to a level that significantly affects plasma iodide. Significant decreases in plasma iodide (i.e., both free and bound) in pigeons were not observed following the exposure to the chlorinated water. Thus it appears that although these disinfectants may affect the dietary level of bound iodide, they do not seem to significantly alter plasma iodide levels.

Other factors that may explain the observed hypothyroid effect of these disinfectants include hypothyroid induction by chloroorganics. Several investigators have shown that chloroorganics are formed when various feeds are mixed with chlorine (*19, 20*). Formation of these compounds may vary with the dietary constituents. Thus, differences in formation rates and species of chloroorganic compounds in the intestinal tract may explain the difference in sensitivity of rabbits and pigeons to chlorinated water. Studies are in progress to determine if chloroorganics are formed in diets mixed with chlorine and if the formed chloroorganics affect the thyroid function.

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## REFERENCES

1. Bercz, J. P., Jones, L., Garner, L., Murray, D., Ludwig, D. A., and Boston, J. subchronic toxicity of chlorine dioxide and related compounds in drinking water in the nonhuman primate. Presented at International Symposium on the Health Effects of Drinking Water Disinfectants and Disinfection By-products, Cincinnati, Ohio, Apr. 21–24, 1981.
2. Revis, N. W., Osborne, T. R., McCauley, P., Bull, R., and Holdsworth, G. The effect of drinking water containing chlorine on cholesterol metabolism in the White Carneau pigeon and New Zealand rabbit. Environ. Health Perspect. (in press).
3. Lister, M. W., and Rosenblum, P. Rates of reaction of hypochlorite ions with sulfite and iodide ions. Can. J. Chem. 41: 3013–3020 (1963).
4. American Public Health Association. Standard Methods for the Examination of Water and Wastewater, 18th ed. A. PHA-AWWA-WPCF, Washington, DC, 1980.
5. Palm, A. T. Methods for the determination in water of the free and combined available chlorine, chlorine dioxide and chlorite, bromine, iodine and ozone, using diethyl-*p*-phenylene diamine (DPD). J. Inst. Water Engr. 21: 537 (1967).
6. Allain, C. C., Poon, L. S., Chan, C. S. G., Richmond, W., and Fu, P. C. Enzymatic determination of total serum cholesterol. Clin. Chem. 20: 470–474 (1974).
7. Sandell, E., and Kolthoff, I. M. Iodine determination. Mikrochim. Acta 1: 9–25 (1937).
8. Keckman, M. M. Iodine in foods. J. Assoc. Off. Anal. Chem. 62: 1045–1049 (1979).
9. AOAC Official Methods of Analysis, 13th ed. AOAC , Arlington, VA, Sect. 7.115, 1980.
10. Snedecor, G. W., and Cochran, W. G. Statistical Methods. Iowa State University, Iowa, 1967.
11. National Academy of Sciences. Nutrient Requirements of Domestic Animals. The National Research Council, 1977.
12. Kritchevsky, D. Influence of thyroid hormones and related compounds on cholesterol biosynthesis and degradation: A review. Metabolism 9: 984 (1960).
13. Gould, R. G. The relationships between thyroid hormones and cholesterol biosynthesis and turnover. In: Hormones and Atherosclerosis (G. Pincus, Ed.), Academic Press Inc., New York, 1959, p. 75.
14. Hazzard, W. R., and Bierman, E. L. Aggravation of broad-B disease (type 3 hyperlipoproteinemia) by hypothyroidism. Arch. Intern. Med. 130: 822–828 (1972).
15. Revis, N. W., Osborne, T. R., Holdsworth, G., and McCauley, P. Effects of chlorinated drinking water on myocardial structure and function in pigeons and rabbits. In: Water Chlorination: Environmental Impact and Health Effects, Vol. 5 (R. L. Jolley, W. A. Brungs, and R. B. Cumming, Eds.), Ann Arbor Science Publishers, Ann Arbor, MI, 1985, pp. 365–371.
16. Revis, N. W., McCauley, P., and Holdsworth, G. The relationship of dietary iodide and drinking water disinfectants to thyroid function in experimental animals. Proc. Natl. Acad. Sci. (in press).
17. Burger, J. D., and Liebhafsky, H. A. Thermodynamic data for

aqueous iodine solutions at various temperatures. Anal. Chem. 45: 600–602 (1973).
18. Palmer, D. A., and Lietzke, M. H. The fifth trihalomethane: Dichloroiodomethane, its stability and occurrences in chlorinated drinking water. In: Water Chlorination: Environmental Impact and Health Effects, Vol. 3 (R. L. Jolley, W. A. Brungs, and R. B. Cumming, Eds.), Ann Arbor Science Publishers, Ann Arbor, MI, 1980, pp. 161–168.
19. Cunningham, H. M., and Lawrence, G. A. Absorption and metabolism of chlorinated fatty acids and triglycerides in rats. Food Cosmet. Toxicol. 15: 101–103 (1979).
20. Cunningham, H. M., and Lawrence, G. A. Effect of chlorinated lipid and protein fractions of cake flour on growth rate and organ weight of rats. Bull. Environ. Contam. Toxicol. 19: 73–79 (1978).